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BREEDING FENUGREEK FOR INDUSTRIAL USES

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# BREEDING FENUGREEK FOR INDUSTRIAL USES

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ABSTRACT

BREEDING FENUGREEK FOR INDUSTRIAL USES

The previous project on fenugreek breeding resulted in two variable populations of fenugreek, a zero-tannin population with reduced color, aroma and taste and a population of normal tannin-containing fenugreek plants. Neither of these populations had been selected for seed yield. Since seed yield is a major factor in determining net returns from fenugreek production, relative to other crops, the objectives were to 1) develop a high yielding cultivar of zero-tannin fenugreek with reduced color, aroma and taste for the functional food and nutraceutical market and 2) develop a high yielding cultivar of normal (tannin-containing) fenugreek for the spice market and other potential markets. Selections from these two populations of fenugreek were evaluated for seed yield over a 3-year period and one line from each population was retained to produce Breeder seed in 2002. CDC Canafen is a new zero-tannin cultivar of fenugreek that yields competitively with the normal (tannin-containing) commercial check cultivar CDC Quatro. CDC Canafen opens up greatly increased uses for fenugreek in the rapidly expanding functional food and nutraceutical industries because of its reduced color, aroma and taste. The second cultivar, CDC Canagreen, produces a markedly higher seed yield than the commercial check cultivar CDC Quatro.

Earlier studies have shown that the cotyledons and the embryo could be manually separated from the remainder, consisting of the gummy endosperm and the inseparable seed coat. The intact fenugreek seed is about 48 % fibre (17 % soluble fibre plus 31 % insoluble fibre). However, the gummy endosperm plus the seed coat fraction consists of about 85 % fibre (38 % soluble fibre plus 47 % insoluble fibre) and 10 % protein. Therefore, 76 % of the total fibre and 95 % of the soluble fibre from the intact seed was concentrated in the gummy endosperm and the inseparable seed coat. The gummy endosperm is almost pure soluble fibre, and is difficult to separate from the seed coat, but this is being done with guar seed to produce guar gum. A detailed study of variability in percentage endosperm plus seed coat was initiated on 23 high yielding non-standard type fenugreek lines from the 2000 and 2001 yield trials plus CDC Quatro and AC Amber fenugreek. In 2000, the percentage endosperm plus seed coat fraction ranged from 45.4 % to 49.7 % of the seed, and was negatively associated with 1000-seed weight ($r = -0.63$ in 2000 and $r = -0.72$ in 2001. That is, the smaller seeds were higher in percentage endosperm plus seed coat than the large seeds. Heritability of percentage endosperm plus seed coat was medium high, but manual separation is not justifiable, even in a breeding program. Some type of mechanical separation of the gummy endosperm is required to make processing of fenugreek seed for fenugreek gum economically feasible.

# BREEDING FENUGREEK FOR INDUSTRIAL USES

A. E. SLINKARD

TECHNICAL REPORT

Fenugreek (Trigonella foenum-graecum L.) is a small-seeded annual legume, currently used primarily as a spice crop. Two cultivars have been released in Canada, AC Amber in 1992 and CDC Quatro in 1995. At the present time, only about 350 acres of fenugreek are planted for seed production each year in western Canada. Recently, interest has also developed in producing fenugreek as an annual forage crop in southern Alberta because of its unique ability to maintain a high protein level all season long.

Two groups of fenugreek are available: one of Indian origin and one of eastern Mediterranean origin. Each group has its own specific strain of Rhizobium. The group originating in the Mediterranean area is best adapted to western Canada, probably because of the similarity in day length between western Canada and the eastern Mediterranean area. Originally, the only inoculum available was specific to the Indian group and fixed little or no nitrogen on the fenugreek plants originating from the eastern Mediterranean. However, this project obtained some inoculum from Russia and it is very effective in fixing nitrogen on the fenugreek plants originating from the eastern Mediterranean. This strain of inoculum is now commercially available from MicroBio Rhizogen (Becker-Underwood) in Saskatoon

The fenugreek seed is rather unique among legume seeds in that it contains little starch or sugar. The carbohydrate fraction consists largely of dietary fibre (44%), which is the major component of interest in this crop. Chemical analysis of the fenugreek seed indicates that it consists of about 44.4 % dietary fibre (32.1 % insoluble and 13.3 % soluble), 36 % protein, 6 % oil, 3.2 % ash, 1.6 % starch and 0.4 % sugars.

Fenugreek is one of the few legumes that has an endosperm. Others include guar (Cyamoposis), locust bean and carob, none of which is adapted to western Canada. The endosperm is a gum that surrounds the cotyledons and embryo, just beneath the seed coat. This gum is a galactomannan, a polysaccharide consisting of galactose (digestible) and mannan (non-digestible). Galactomannans are a source of soluble fibre and increase viscosity when dissolved in an aqueous solution. Guar gum is widely used as an adhesive, emulsifier and as a thickening agent in various foods such as soups, stews, ice cream and sherbet. Guar gum galactomannan consists of a chain of one molecule of galactose and two molecules of mannan, etc. whereas the fenugreek gum galactomannan consists of a chain of one molecule of galactose and one molecule of mannan, etc. Both of these galactomannans are considered soluble fibre and comprise most of the endosperm in seeds of these species. Clinical trials have established that defatted fenugreek flour is effective in reducing serum glucose levels in type II, non-insulin dependent diabetes, has hypocholesteromic effects and functions as an appetite suppressant.

Pure mannan is completely indigestible and is insoluble in water, but solubility increases with the addition of galactose to the mannan: the higher the galactose ratio, the higher the solubility. Thus, fenugreek galactomannan (50% galactose) has higher solubility in water than guar galactomannan (33% galactose). Both guar galactomannan and fenugreek galactomannan act as an emulsifier and stabilizer in oil-water mixtures and have many potential applications in the food industry and elsewhere as a thickening agent. The challenge is to produce and process a colorless, tasteless and odorless product at prices competitive with the industry standard, guar gum.

However, the taste, color, smell and fat level are too high in the unprocessed fenugreek flour, and must be removed by expensive processing. If the level of one or more of these deleterious factors could be reduced or eliminated genetically, the processing costs would be reduced and the product would be more competitive in the market place. Likewise, the development of higher yielding cultivars of fenugreek for Saskatchewan would make Saskatchewan more competitive in the rapidly developing fenugreek industry.

## FENUGREEK RESEARCH AT THE CROP DEVELOPMENT CENTRE PRIOR TO INITIATION OF THIS PROJECT

In 1989, Ferdinand Kiehn, Morden Research Station, Agriculture and Agri-Food Canada, entered three fenugreek lines in the New Crops Co-operative trials. This co-operative test was grown at over 20 site-years in 1989 to 1991, including three years at Saskatoon. NC 109-1 was the highest yielding of these three entries. It was a bulk increase of PI 138,687 (Iran) from the USDA Plant Introduction Service and was released exclusively to G. H. Schweitzer Enterprises, Eston SK in 1992 as the cultivar AC Amber.

### A. AGRONOMIC STUDIES

In 1990 and 1991, the CDC conducted a series of agronomic trials on factors affecting seed yield in fenugreek, using the seed of NC109-1 harvested from the 1989 Co-operative Fenugreek Trials plus several seed lots contributed by Gary Schweitzer of Eston SK. These trials included: 1) Date of Seeding (early May seeding resulted in higher seed yields than late May seeding), 2) Seeding Depth (seed yields decreased when planted 5 cm deep or deeper), 3) Row Spacing (30-cm row spacing produced seed yields similar to those with a 10, 20 or 40-cm row spacing), 4) Seeding Rate (seeding rates of 90, 135 or 180 seeds/square metre [= 8, 12 or 16 seeds/square foot, respectively] did not differ much in seed yield and the intermediate rate [135 seeds/square metre = 12 seeds/square foot provided some margin for error] and was the suggested rate of seeding), 5) Phosphorus Rate (80 kg/ha of phosphate had little effect on seed yield), 6) Inoculant Test (inoculant from the US had little, if any, effect on seed yield, whereas inoculant from Russia (MicroBio Rhizogen) consistently resulted in slight increases in seed yield), and 7) Irrigation Test (irrigation increased seed yield and dry matter yields in a dry year, but had little effect on seed yield, but increased dry matter yields and delayed maturity in a year with somewhat normal rainfall). A Desiccation Trial was conducted in 1997, comparing the effects of KI, KCl and Reglone (diquat). No differences in seed yield were noted,

presumably due to the fact that the fenugreek pods do not shatter readily, and the crop can be combined directly. A companion cropping study indicated that fenugreek was more competitive than coriander.

Several herbicide-screening trials were conducted between 1991 and 1996. Results indicated that fenugreek is susceptible to metribuzin (post-emergent), bentazon, 2,4-DB, MCPA/MCPB, and propanil, but that trifluralin (Treflan), ethalfluralin (Edge), cyanazine, pendimethalin and low rates of imazethapyr (Pursuit) may provide acceptable weed control without seriously damaging seed production of fenugreek. Fenugreek had good tolerance to all graminicides tested. In 1996, fourteen zero-tannin lines of fenugreek were evaluated for tolerance to the herbicide bentazon and all were damaged, but recovered to near normal. This indicates that the zero-tannin trait did not predispose the fenugreek cultivars to any more herbicide damage than the normal tannin-containing cultivar CDC Quatro fenugreek, at least to bentazon.

B. BREEDING STUDIES

In 1990, the USDA Fenugreek Collection (143 Plant Introduction [PI] lines) was increased in short rows at Saskatoon.

In 1991, 71 of the PI lines of fenugreek that produced enough seed plus AC Amber were grown in preliminary yield trials with three replications at Saskatoon. Data were collected on seed yield, percentage hull, days to flower, days to mature, plant height, 1000-seed weight, percentage hull, percentage cotyledons, percentage root plus shoot, percentage protein, percentage total digestible fibre, percentage soluble fibre and soluble fibre as percentage of total fibre on 24 of the 72 lines tested. Only 24 lines were studied in detail because these determinations are very time consuming and expensive. The highest yielding entry was PI 143504, followed by AC Amber. PI 143504 had a lower percentage of both protein and cotyledons, but heavier (larger) seed, and a higher percentage of endosperm plus seed coat, root plus shoot, total digestible fibre, soluble fibre and soluble fibre as percentage of total dietary fibre than AC Amber fenugreek.

Also, in 1991, five cultivars of fenugreek were grown in a replicated regional yield trial at Saskatoon and Bounty. Data were collected on seed yield, other agronomic traits and several oil components. The seeds of these five cultivars contained an average of 4.49 % oil which consisted of 13.9 % palmitic acid, 4.70 % stearic acid, 15.3 % oleic acid, 37.7% linoleic and 24.4 % linolenic acid. Since the percentage oil was so low and the fatty acid composition was in agreement with means in the literature (except for the low percentage linolenic reported here [24.4 % vs. over 35 % reported in the literature]), no further data were collected on oil in the fenugreek seed. The fenugreek seed also contains about 0.3 % saponin.

In 1992, thirty-five of the highest yielding PI lines (out of 71) were grown in a replicated yield trial (6 x 6 lattice with six replications) at Saskatoon. A bulk increase of PI 143504 was also entered in the 1992 Co-operative Fenugreek Trials. It was among the highest

yielding line in both tests. Accordingly, it was retained in the Co-operative Fenugreek Trial for second year testing in 1993, and for third-year testing in 1994, where it had the highest seed yield, based on the three-year average.

Concurrently, in anticipation of its potential release, 100 random plants from the bulk seed increase of PI 143504 were selected in the fall of 1993. Seeds from these 100 selected plants were grown in plant rows in 1994, selected for trueness- to - type and the most uniform and phenotypically similar plant rows were harvested in bulk to produce Breeder seed of CDC Quatro fenugreek which was tendered as an exclusive release in 1995. The tender was awarded to Emerald Seed Products, Regina SK. NOTE: No replicated yield trials were conducted on the progenies of these selections, since the lines were bulked solely on the basis of seed yield of the parental PI line and phenotypic similarity of the bulked lines.

. An off-type with a zero-tannin seed coat was identified in 1991 and crosses made with it to determine its mode of inheritance. In addition, crosses were made between various accessions of fenugreek and the mode of inheritance of nine isozyme polymorphisms (ACO-2, AMY-2, AAT, EST-1, EST-2, ESR-3, GDH, PRX-3 and TPI) was determined in the second filial generation. All nine isozymes showed monogenic control and Mendelian inheritance with codominant expression of the alleles. The quaternary structure of AAT was dimeric and the remaining isozymes were monomeric. Monogenic inheritance was also observed for four morphological traits (cotyledon colour - green vs yellow, pubescence - glabrous vs pubescent, single vs double pods/node, and presence vs absence of anthocyanins or tannin precursors). Green cotyledon was dominant (?) to yellow, glabrous foliage was dominant to pubescent, double pod was dominant to single pod/node and tannin-containing seed coat was dominant to zero tannin seed coat. One linkage group was identified: Tpi-1 - 25.3 cMorgans - Est-2 - 2.02 cMorgans - Yc =yellow cotyledons - 3.87 cMorgans -Est-3. Close linkage was also found between Prx-3 and Amy-2 in one population (identical segregation patterns). These results are presented in detail in Harikumar Nair's Masters' thesis in 1994.

In 1994, fifty individual F-2-derived F-4 plants with zero-tannin seed coats were selected from crosses involving the zero-tannin trait. These were increased in 1995.

In 1996, fourteen of the most promising zero-tannin fenugreek families plus AC Amber and CDC Quatro as the normal (tannin-containing) check cultivars were entered in a preliminary yield trial at Saskatoon. Data were collected on seed yield, days to flower, days to mature, plant height and 1000- seed weight. In general, the zero-tannin lines were agronomically similar to the normal (tannin-containing) cultivars in all traits except seed yield, which was lower than that of CDC Quatro, except for four families.

In this 1996 yield trial detailed data were also collected on percentage of protein, ash, crude fat, soluble dietary fibre, soluble fibre (as percentage of total fibre) and total dietary fibre. The zero-tannin families averaged higher in percentage of protein, soluble dietary protein and soluble fibre (as percentage of total dietary fibre), but were lower in percentage total dietary fibre than the normal (tannin-containing) check cultivars. Further

analysis of the fenugreek seed indicated that the gummy endosperm plus seed coat fraction consists of about 85 % total dietary fibre (38 % soluble fibre plus 47 % insoluble fibre, i.e., the seed coat plus the gummy endosperm contain about 95 % of the soluble fibre in the fenugreek seed). The gummy endosperm is almost pure soluble fibre. The problem then becomes a matter of separating the seed coat from the gummy endosperm, which is almost pure soluble fibre. An inexpensive and efficient method for this process would make fenugreek competitive with guar gum as an adhesive, thickening agent and emulsifier.

At this time, funding for the original project on breeding fenugreek expired. Subsequently, the decision was made to seek further funding and to initiate a new cycle of selection with emphasis on seed yield. Accordingly, in 1998, about 100 plants from within CDC Quatro (a bulk increase of PI 143504) fenugreek and about 300 zero-tannin plants from the highest yielding segregating family were randomly selected and increased in anticipation of renewed funding. New funding was approved in February 1999 to complete the evaluation of these two groups of fenugreek selections.

START OF NEW PROJECT

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BREEDING FENUGREEK FOR INDUSTRIAL USES

OBJECTIVES:

The objectives of this project were to develop 1) an early maturing, high yielding cultivar of zero-tannin fenugreek that is high in soluble fibre with reduced taste, aroma and color for use in the rapidly expanding functional food and nutraceutical markets, and 2) an early maturing, high yielding cultivar of normal (tannin-containing) fenugreek, thereby establishing the basis for Saskatchewan to improve on its role as the Canadian leader in the production, processing and marketing of fenugreek.

MATERIALS AND METHODS:

In 1998, about 100 individual plants were randomly selected from within CDC Quatro (a bulk increase of PI 143504) fenugreek and about 300 individual plants were randomly selected from the highest yielding segregating family of zero-tannin fenugreek.

In 1999, these 400 selected plants were increased in mini-plots.

In 2000, 279 of the highest yielding zero-tannin lines were planted in a two-replication preliminary yield trial and agronomic data were collected on seed yield, date of first flower, plant height and 1000-seed-weight. Several zero-tannin lines had a higher seed yield than the commercial check cultivar CDC Quatro fenugreek.

In 2001, 63 of the highest yielding zero-tannin lines (out of 279 tested in 2000) were planted in a replicated (3x) yield trial at Saskatoon. These included the 23 highest yielding zero-tannin lines, which were tested for 1000-seed weight and percentage endosperm plus seed coat in both the 2000 and 2001 yield trials. Detailed data were collected on two replications for 1000-seed weight and percentage endosperm plus seed coat of these lines. This is a very labour-intensive operation and was not completed until the summer of 2002.

The fenugreek seeds were tempered so that the cotyledons plus embryo could be manually separated from the endosperm plus the inseparable seed coat. Fifty seeds were placed in a small plastic petri dish (in duplicate) and placed on a platform over water in a sealed plastic pail. Seed of most lines of fenugreek was softened enough after 96 hours of tempering so that the seed could be cut in half with a scalpel and the cotyledons plus embryo fraction separated from the endosperm plus seed coat fraction. At least 60% of the seeds (30) was cut for each sample. Some fenugreek lines had a high hard seed content and a second sample of 50 seeds was tempered in order to get the minimum of 30 cut seeds per test. The bulked endosperm plus seed coat fraction was separated from the bulked cotyledon plus embryo fraction, dried overnight at room temperature, weighed separately and percentage of endosperm plus seed coat calculated. This was a time consuming task and was done on seeds from two replications of this trial in both 2000 and 2001 in order to determine the range of variability and the repeatability (broad sense heritability) of this trait.

Also, in 2000, detailed data were collected on seed coat colour, percentage oil, constituent fatty acids in the oil and percentage diosgenin of 30 zero-tannin fenugreek lines and 8 normal (tannin-containing) fenugreek lines. Early maturing spaced plants did not consistently produce early maturing mini-plots. Thus, individual plant selection for maturity in spaced plants of an indeterminate flowering legume, such as fenugreek, is a waste of time, unless large differences in maturity are present. No differences were noted between the zero-tannin lines and the normal (tannin-containing) lines, except for seed coat colour, which was readily evident without precise measurements. Consequently, no additional data were collected on any of these traits except for seed yield, 1000-seed weight and percentage endosperm plus seed coat.

Also, in 2000, about 100 plants of CDC Quatro fenugreek were increased in miniplots and the 50 highest yielding were entered into replicated yield trials in 2001 and the eight highest yielding lines were retested in 2002.

In 2001, 63 of the highest yielding zero-tannin lines (out of 279 tested in 2000) were planted in a replicated (3x) yield trial at Saskatoon. These included the 23 highest yielding zero-tannin lines, which were tested for 1000-seed weight and percentage endosperm plus seed coat in 2000 and repeated in 2001.

In 2002, the highest yielding normal (tannin-containing) fenugreek line (MP36 = CDC Canagreen) and the highest yielding zero tannin line (LF5 = CDC Canafen) were planted in Breeder seed plots. Selection was based on their high average seed yield in 2000 and 2001. In addition, the eight highest yielding normal (tannin-containing) lines of fenugreek and the 28 highest yielding zero-tannin lines of fenugreek were established in a 5-replication trial. Data were collected on seed yield in 5 replications and on 1000-seed weight in two replications

RESULTS:

Seed yield trials and Breeder seed plots in 2002:

One zero-tannin line of fenugreek (LF-5 = CDC Canafen) was planted in a Breeder seed plot in 2002. Twenty-right zero-tannin and eight normal (tannin-containing) lines of fenugreek, including CDC Canafen, CDC Canagreen and CDC Quatro, were established in a replicated seed yield trial at Saskatoon to confirm the choice of line LF-5 as the best line for the Breeder seed plot of the new zero-tannin cultivar of fenugreek, CDC Canafen, and to confirm that MP36 was the best line for the Breeder seed plot of the new normal (tannin-containing) cultivar of fenugreek, CDC Canagreen. The summer of 2002 was unusually dry and the seed yields were low. Nevertheless, CDC Canafen fenugreek yielded 626 kg/ha or 73 % of CDC Quatro fenugreek. However, it yielded 4 % more than CDC Quatro over the three years of testing at Saskatoon (Table 1), i.e. seed yield of CDC Canafen fenugreek is comparable with that of CDC Quatro fenugreek. Both CDC Canafen zero-tannin fenugreek and CDC Canagreen normal (tannin-containing) fenugreek produced the highest seed yield in their respective classes, and higher than the commercial check cultivar CDC Quatro fenugreek, based on their average yields in tested over 3 and 2 years, respectively (Tables 1 and 2).

Table 1. Three-year seed yields of CDC Quatro and CDC Canafen fenugreek at Saskatoon, 2000-2002.

| Cultivar | 2000 | 2001 | 2002 | Mean |
|---|---|---|---|---|
| | Kilograms per hectare | | | |
| CDC Quatro | 1886 | 1750 | 858 | 1498 |
| CDC Canafen | 2157 | 1908 | 626 | 1563 |

Seed of CDC Quatro and CDC Canagreen fenugreek, but not CDC Canafen fenugreek, was also supplied to AAFC at Lethbridge for seed yield and forage yield comparisons in 2002. A comparison of the seed yields of CDC Canagreen and CDC Quatro fenugreek at Saskatoon in 2001 and 2002 plus Lethbridge in 2002 is presented in Table 2. In this comparison, seed yield of CDC Canagreen fenugreek was 28 % higher than that of CDC Quatro fenugreek. Thus, seed yield of CDC Canagreen fenugreek normal (tannin-containing) fenugreek, a reselection, based on replicated progeny trials, from within the parental PI line for CDC Quatro was higher than a bulk seed increase of the original parental PI line.

Table 2. Two-year seed yields of CDC Quatro and CDC Canagreen fenugreek at Saskatoon, 2001-2002 and Lethbridge in 2002.

| Cultivar | 2001 | 2002 | Lethbridge (2002) | Mean |
|---|---|---|---|---|
| CDC Quatro | 1678 | 858 | 442 | 993 |
| CDC Canagreen | 2205 | 972 | 656 | 1278 |

One normal (tannin-containing) line of fenugreek (MP-36 = CDC Canagreen) was planted in a Breeder seed plot in 2002. A comparison of the seed yield and forage yield of CDC Canagreen and CDC Quatro fenugreek is presented in Tables 2,3 and 4, and indicates that CDC Canafen normal (tannin-containing) fenugreek is similar to CDC Quatro in most agronomic traits, except seed yield, where it is higher yielding (Table 2).

Table 3. Summary of agronomic traits for CDC Quatro, CDC Canafen and CDC Canagreen fenugreek at Saskatoon, 2000-2002.

| Cultivar | Height (cm) | 1000-seed weight (g) | Endosperm and seed coat (%) |
|---|---|---|---|
| CDC Quatro | 40 | 19.0 | 49.7 |
| CDC Canafen | 33 | 22.8 | 44.8 |
| CDC Canagreen | 42 | 18.5 | 49(?) |

Table 4. Forage yield of several fenugreek lines from Saskatoon and Lethbridge at Lethbridge in 2002.

| Line | Forage yield (kg dry matter/ha) |
|---|---|
| L3308 | 4872 |
| AC Amber | 4573 |
| L3312 | 4578 |
| MP-30 | 4280 |
| CDC Quatro | 3872 |
| MP-36 | 3793 |
| LSD (0.05) | 1741 |

Percentage endosperm plus seed coat and 1000-seed weight in 23 lines of zero-tannin fenugreek, 2000 and 2001

Data on 1000-seed weight and percentage endosperm plus seed coat for 23 lines of zero-tannin fenugreek in 2000 and 2001 is presented in Table 5. In 2000, percentage endosperm plus seed coat ranged from 42.9% to 48.2 % (49.7 % for CDC Quatro). This range is large enough to make meaningful progress, provided it is repeatable, i.e., largely due to genetic variation. Accordingly, these lines were retested in the 2001 yield trial and the results are presented in Table 5.

Table 5. Average seed yield, 1000-seed weight and percentage endosperm plus seed coat for 23 zero-tannin fenugreek lines, including CDC Canafen and the two normal (tannin-containing) check cultivars of fenugreek, AC Amber and CDC Quatro, from the 2000 and 2001 yield trials at Saskatoon.

| Line | 2000-2001 ave. yield (kg/ha) | 1000-seed weight (g) 2000 | 1000-seed weight (g) 2001 | Endosperm plus seed coat %) 2000 | Endosperm plus seed coat %) 2001 |
|---|---|---|---|---|---|
| CDC Quatro | 1818 | 15.4 | 15.5 | 49.7 | 45.3 |
| LF -5 (Canafen) | 2032 | 16.7 | 18.5 | 44.8 | 41.1 |
| EF-54 | 1454 | 16.2 | 18.2 | 48.2 | 42.9 |
| EF-27 | 1706 | 17.1 | 19.3 | 47.4 | 41.0 |
| EF-10 | 1693 | 15.7 | 19.1 | 47.1 | 41.5 |
| EF-41 | 1920 | 15.5 | 18.4 | 47.1 | 41.6 |
| EF-35 | 1638 | 16.8 | 17.8 | 46.8 | 43.6 |
| EF-49 | 1668 | 17.9 | 18.0 | 46.5 | 41.2 |
| EF -46 | 1624 | 17.2 | 19.7 | 46.4 | 41.9 |
| LF-71 | 1784 | 17.1 | 20.2 | 46.2 | 41.2 |
| EF -19 | 1676 | 17.2 | 18.8 | 45.5 | 41.7 |
| EF-75 | 1801 | 17.9 | 18.5 | 45.4 | 42.4 |
| EF- 25 | 1672 | 16.8 | 19.0 | 45.2 | 41.9 |
| EF-69 | 1620 | 16.8 | 19.7 | 44.8 | 41.6 |
| EF-2 | 1634 | 17.1 | 18.1 | 44.7 | 40.3 |
| EF -6 | 1613 | 16.5 | 19.6 | 44.6 | 40.7 |
| LF 9 | 1808 | 17.2 | 19.0 | 44.6 | 43.2 |
| AC AMBER | 1992 | 15.8 | 15.9 | 44.3 | 41.2 |
| EF 51 | 1666 | 18.5 | 18.8 | 44.0 | 39.8 |
| EF 5 | 1722 | 17.6 | 19.1 | 44.0 | 38.5 |
| LF -41 | 1785 | 18.0 | 18.5 | 43.8 | 41.0 |
| EF -21 | 1425 | 17.3 | 19.2 | 43.7 | 38.6 |
| EF-16 | 1609 | 18.0 | 18.6 | 43.5 | 40.2 |
| EF-18 | 1522 | 18.2 | 19.5 | 43.2 | 40.5 |
| EF-12 | 1643 | 17.3 | 17.5 | 42.9 | 41.1 |
| MEAN | 1701 | 17.0 | 18.5 | 45.0 | 41.4 |
| LSD (0.05) | --- | 1.7 | | 1.9 | |

Correlations between the 2000 data and the 2001 data were calculated to determine their repeatability. The correlation between parents (2000 data) and progeny (2001 data) also provides an estimate of broad sense heritability in terms of "standard units". The correlation coefficient squared (r-squared) is termed the "coefficient of determination" and gives the percentage variation in the progeny (2001 data) that is due to variation in the parents (2000 data).

The seed coats of the non-standard type fenugreek were whiter (higher L values), redder (higher a values) and yellower (higher b values) than seed coats of the standard type fenugreek. Seed weight was negatively correlated ($r = -0.63$ in 2000 and $r = -0.72$ in 2001) with percentage endosperm plus seed coat, i.e., the smaller seeds had a higher percentage endosperm plus seed coat than the larger seeds. The tannin-containing check cultivars had a higher percentage endosperm plus seed coat than the zero-tannin lines because they had a thicker, heavier seed coat.

"Broad sense" heritability in standard units for percentage endosperm plus seed coat was estimated by the correlation coefficient between the years 2000 and 2001 and was 0.567 (significant) for percentage endosperm plus seed coat for the 23 zero-tannin lines of fenugreek. Thus, one can successfully select for this trait. This was further confirmed by the medium high coefficient of determination (r squared = 32 %). However, selection for percentage endosperm plus seed coat is not worth while due to the expense involved and would have to be restricted to within a zero-tannin population or within a normal (tannin-containing population

"Broad sense "heritability in standard units was estimated for 1000-seed weight by the correlation coefficient between the years 2000 and 2001 and was 0.064 (non-significant) for 1000-seed weight. This is contrary to reports in other crops and is likely due to the limited genetic variability (small range) in 1000-seed weight among these 23 lines of zero-tannin fenugreek, all selected from one high yielding F-2 derived family.

Summary:

1. Breeder seed of CDC Canafen zero-tannin fenugreek was developed and it produces seed yields competitive with the commercial check cultivar, CDC Quatro fenugreek.

2. Breeder seed of CDC Canafen normal (tannin-containing) fenugreek was developed and it produces seed yields markedly higher than the commercial check cultivar CDC Quatro fenugreek.

3. Selection for differences in maturity among individual spaced plants of an indeterminate plant, such as fenugreek, was ineffective, unless very large differences are present.

4. Selection for 1000-seed weight within F 2-derived families will be of limited effectiveness because of the limited genetic variability for this otherwise highly heritable trait within each F2-derived family.

5. Percentage endosperm plus seed coat is a moderately heritable trait, but it is too expensive to measure and improvement is slow, partially due to the negative correlation with seed weight and the consistent differences between the zero-tannin lines and the tannin-containing lines. The tannin containing lines had a thicker, heavier seed coat than the zero-tannin lines and, thus they had a higher percentage endosperm plus seed coat.

6. An effective, efficient and low cost processing method must be developed to produce pure gummy endosperm, if fenugreek gum is to compete effectively with guar gum in functional food and nutraceutical markets.

Information of benefit to the Saskatchewan, due either to limited variability or an agri-food industry

A zero-tannin cultivar of fenugreek, such as CDC Canafen, has a reduced color, aroma and taste, relative to other fenugreek cultivars. This will open up increased opportunities for Saskatchewan fenugreek producers in the rapidly developing market for fenugreek seed in the food and nutraceutical industries. The development of a niche market processing industry for this rapidly expanding industry is currently in progress. The seed yield of CDC Canafen zero-tannin fenugreek is competitive with that of the commercial tannin-containing cultivar CDC Quatro. Thus, a new cultivar with this valuable trait was developed without any reduction in seed yield.

A normal (tannin-containing cultivar of fenugreek, CDC Canagreen, was developed by reselection from within the original PI line, which was bulked to produce Breeder seed

for CDC Quatro. The individual parental lines that were bulked to produce the Breeder seed of CDC Quatro fenugreek were not evaluated by seed yield. Thus, adequate genetic variability for seed yield remained within the population so that testing of random lines for seed yield in replicated yield trials resulted in the development of a line with a marked increase in seed yield over the original cultivar, CDC Quatro. This improved cultivar will serve to make Saskatchewan more competitive in the production of fenugreek for the spice industry and other uses.

Potential for commercialization:

The availability of a high yielding cultivar of fenugreek with reduced color, aroma and taste (CDC Canafen zero-tannin fenugreek) will find wide application in the rapidly expanding functional food and nutraceutical industries. Fenugreek endosperm is almost pure soluble fibre. If an effective, efficient and low cost method could be developed to separate the gummy endosperm from the seed coat to produce pure gummy endosperm, fenugreek gum has the potential to replace the industry standard guar gum in many food and nutraceutical uses. Methods have been developed for guar, but they are trade secrets. The economics of fenugreek production and processing in Saskatchewan will be greatly improved with this development. Saskatchewan already is the major provincial producer and processor of fenugreek. The area devoted to fenugreek production could easily increase from an estimated 350 acres in 2001 to 3500 acres by 2010. The impending construction of a commercial scale fenugreek processing plant in Saskatchewan will aid in rural development and help improve the overall farm economy.

Project materials:

1. Breeder seed of CDC Canafen zero-tannin fenugreek with seed yield equivalent to the check cultivar CDC Quatro fenugreek
2. Breeder seed of CDC Canagreen fenugreek with a marked increase in seed yield relative to the commercial check cultivar CDC Quatro.

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Lori Jackson -laboratory technician
Ghasem Bharani -field technician

Equipment:

None

Expense statement:

In process

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